ENDAMOEBA BUCCALIS

II. ITS REACTIONS AND FOOD-TAKING

NADINE NOWLIN
University of Kansas

The material for the present paper was collected from the same source as that of the previous study — a single host and a single point of infection, an upper premolar tooth. For details of preparation of material and method of study see Nowlin (1917).

MORPHOLOGY AND BEHAVIOR

The size of the living trophozoite found in smears varies from about 12 to 40μ when in a spherical condition, and the latter may elongate to 80μ. The amoebae most frequently encountered are from 25 to 35μ, constantly forming pseudopodia if it be no more than shoving out a slight rim on one side and withdrawing it. A specimen just out of the mouth is usually active unless it has been chilled or is forming a cyst. It is not necessarily progressing, but can be seen to change shape rapidly by sending out blisters of ectosarc one-third its entire size. One of these rapidly melts into another and will continue to do so unabated for an hour or more if conditions of warmth and moisture are favorable. A group of twenty amoebae massed into a clump were observed vigorously crawling over each other for more than an hour. Food material was the suspected stimulus for such motion, but when they finally separated from a sudden cooling of the slide, no food was visible. They might have exhausted any supply, but more probably they hung together thru positive thigmotaxis.

During progressive movement the endamoeba is an interesting example of the highest development of pseudopodial motion. The animal is elongated to about the proportions of a thumb, and clearly differentiated as to ends. Forward is a clear protrusion of ectoplasm nearly half the length of the body, and rounded out like a bag. At the posterior end is a knob (Fig. 1*a*), which, if torn from an attachment will have little papillae or beads on the periphery (Fig. 1*c*). These threads of attachment enable an amoeba to carry about with it great masses of leukocytes and debris. It exhibits in this most remarkable strength for a one-celled organism. One amoeba can break up a large solid-looking mass of leukocytes by crowding into them. It can load up with a great cargo of cells and tartar which it carries over the slide as long as one has the patience to watch; nor does the load seem to impede its progress.

The very active movements of *Endamoeba buccalis* in smear preparations suggest great possibilities of damage in the gum by purely mechanical processes, tho this is by no means their full capacity for mischief, as will be seen under methods of food-getting.

Reactions to Light and Heat.—Endamoeba buccalis exhibits a much more marked positive thigmotaxis than do free living amoebae. To leave deliberately a clump of leukocytes to which it has been attached and venture into the open requires several attempts. In one case under observation there was a gap a little greater than the width of the endamoeba's body to be crossed before more leukocytes could be reached. The animal extended itself probably twenty times toward the mass, before it relinquished hold, and even then only after it had succeeded by an extraordinary stretch in touching the other side. These parasites have the capacity of stringing a pseudopod to a length of five or six times their body diameter. Attached thus an animal may

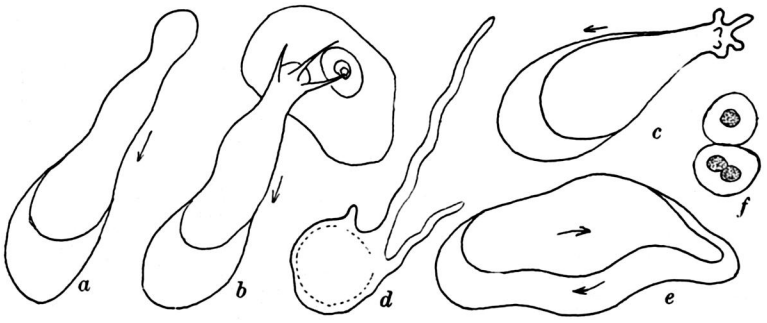

Fig. 1.—*Endamoeba buccalis* in movement. For details see text.

wander into a clear field, breaking the thread only when it comes in contact with a solid, or settles down for attachment to the slide. Many amoebae appearing free are found to be thus attached if the light is properly adjusted. Again the pseudopodia may assume a perfect corkscrew appearance, attached to the spherical body of the parasite and moving in various directions like waving tentacles (Fig. 1*d*).

The few experiments performed to test their sensitiveness to light suggest that they have lost this quality. No difference in behavior could be detected in very bright light and in reduced rays. They are very sensitive to heat, however, and where that is combined with light, as is often the case, there is marked reaction, as shown by increase of activity so long as that temperature is kept around body warmth.

No contractile vacuole is present, and only twice in the living specimens have anything like nuclei been observed. One morning every living amoeba in the smears, and many were made, showed a single red spot about the size of the nucleus of stained specimens. The host

had gargled with glycothymoline and that was suspected as the staining agent, but it could never be demonstrated again either by direct application or by gargling. Under one other condition a spot resembling the nucleus showed up uniformly in many living forms which had been kept sealed for six hours under a cover-slip. These endamoebae assumed an encysted form without completing it with the outer wall, lost all their vacuoles, and showed a single dense spot slightly to one side of the center. This may have been a nucleus.

Food and Food Habits.—The adult or mature trophozoite form has from one to twenty food vacuoles. The usual appearance is three or four of these, each about one-fourth the diameter of the amoeba, and many smaller ones. In life, the contents of these large vacuoles are homogeneous spherical masses, having a greenish gray color, and showing only a single mass to a vacuole (Figs. 2*a* and *b*). In stained specimens also the food vacuole contents are solid masses, varying

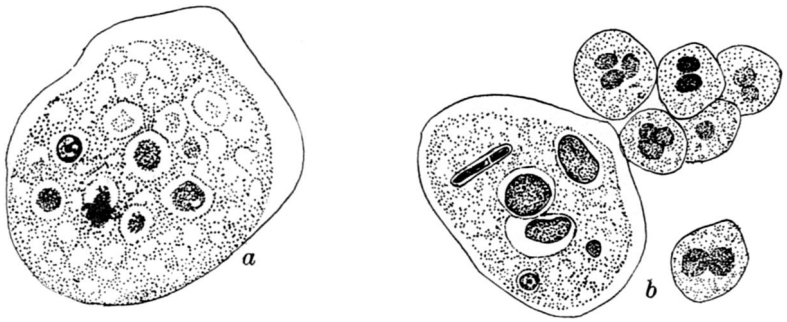

Fig. 2.—*a, Endamoeba buccalis* stained with Dobell to show nucleus. *b,* Stained as above to show food vacuoles and attached leucocytes.

only in intensity of coloring, and showing never any resemblance to leukocytes. They appear more as globules of fluid. No endamoebae under observation during this study, though they have been watched carefully for this, have been seen to ingest leukocytes, red blood cells, as described by other writers, notably Smith and Barrett (1915), or anything solid except bacteria, and these in small numbers. That they never do this, I cannot say definitely, but I believe the following behavior may have been interpreted as ingestion.

The endamoebae frequently attach themselves to leukocytes (Fig. 2*b*). I have seen them creep over the leukocyte and practically surround it until it looked quite like ingestion. A leukocyte will even become sufficiently incorporated to be taken away with the endamoeba for a short time, but if watched sufficiently long the leukocyte is invariably left on the slide, apparently unharmed. I have seen other clear refractive bodies taken in like this and soon discharged, never becoming real food vacuoles.

SUMMARY

I conclude from my observations that *Endamoeba buccalis* absorbs its food mainly, taking in by osmosis the fluids of leukocytes or other media on which it rests, stores these colloidal substances in vacuoles, and by secretion of its own enzymes assimilates these as needed.

The reasons against believing that large food vacuoles are ingested leukocytes may be summarized thus:

(1) There is never but one body to a vacuole, while most leukocytes have one to three nuclei.

(2) There is never any granular area around the vacuolar inclusions, as would be the case if the cytoplasm of a leukocyte were ingested.

(3) Leukocytes have been surrounded by amoebae, but never ingested, according to my observations.

(4) The whole system of vacuoles can vanish from an *Endamoeba buccalis* exposed to unfavorable conditions, sooner than would be possible if these were solid inclusions; moreover, the leukocytes outside the endamoeba are left intact.

This method of food-getting by absorption would explain the shrinkage of gums where *Endamoeba buccalis* is present. I have seen no evidence of their penetrating epithelial cells, but there is abundant evidence that they draw supplies by applying themselves to the surface of tissues and by crowding between them (Fig. 2).

REFERENCES CITED

Nowlin, Nadine. 1917. *Endamoeba buccalis.* I. Its Multiplication and Periodicity. Jour. Parasit., 3:143-149; 1 fig.

Smith, A. J., and Barrett, M. T. 1915. The Parasite of Oral Endamoebiasis —*Endamoeba gingivalis* (Gros). Jour. Parasit., 1:159-174; 1 pl.